

Chromatographic Separation of Melanin Granules.

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Separation of cytoplasmic particulate constituents from cells has heretofore been accomplished principally by differential centrifugation which exploits the size and specific gravity of the particles. A chromatographic system has recently been obtained which permits the separation of some of the cytoplasmic constituents from mouse melanomas by exploiting still other properties of the particulates, presumably their surface characteristics as expressed by their different adsorption affinities. By utilizing the adsorption and elution reversibility of some of the particulates, melanin granules have been separated from the Cloudman S91 and the Harding-Passey mouse melanomas in a highly active state with respect to succinoxidase, cytochrome oxidase and dopa oxidase enzyme activities. Relatively mild adsorption systems, varying between physiological saline and distilled water as the solvent extremes, and involving diatomaceous silica (Celite) columns, have been employed in order to

avoid enzymic inactivation. No gross morphologic changes have been observed in the granules whether separated chromatographically or centrifugally.

A systematic study of the effects of various ions at graded concentrations in adsorbing and eluting melanin granules is under investigation, especially with reference to the possibility of obtaining granules of varying composition, either pre-existing in the tumor or induced by column passage.

The adaptation of chromatography to subcellular particulates provides a new approach to the problem of separating from the cell morphologically similar units possessing different physical or chemical surfaces.

BIBLIOGRAPHY.

1. RILEY, VERNON T. Application of chromatography to segregation studies of the agent of chicken tumor I (Rous sarcoma virus). *Science*, 107: 573-575, 1948.
2. RILEY, VERNON T., HESSELBACH, MARIE L., FIALA, SILVIO, WOODS, M. W. AND BURK, DEAN. Application of chromatography to the separation of subcellular, enzymatically active granules. *Science*, 109: 361-364, 1949.
3. RILEY, VERNON AND WOODS, M. W. A short column procedure for separating cytoplasmic components from normal and tumor tissue. *Proc. Soc. Exp. Biol. & Med.*, 73:1, 92-95, 1950.